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SHORTER ARTICLES AND DISCUSSION

IS THE CHANGE IN THE SEX-RATIO OF THE FROG, THAT IS AFFECTED BY EXTERNAL AGENTS, DUE TO PARTIAL FERTILIZATION?

IN a review in this journal (XLV, 1911) of certain experiments by Kuschakewitsch¹ on frogs' eggs in which by delaying fertilization for 89 hours he obtained 100 per cent. of males, I pointed out that unless more than half of the eggs were fertilized the *interpretation* of the 100 per cent. ratio might be misleading. For should the delay act more injuriously on one kind of egg than on the other, assuming two kinds to exist, the result might mean only selective destruction by an external agent rather than a change in sex of the eggs. I found no explicit statement in the section of Kuschakewitsch's paper dealing with these results to show whether or not all of the eggs had been fertilized, but in a recent rejoinder² to my review Kuschakewitsch points out that he had stated that practically all of the eggs ("so gut wie alle Eier") were fertilized and developed. This information is given in an *appendix* which I had overlooked. His statement completely sets aside the possibility of the suggestions that I made, but leaves the explanation of his results as obscure as before.

The details of the principal experiment and of some of the others are of interest. A pair of copulating frogs were caught at 12:00, midday, May 31. The female began to lay at once. At 6:00 P.M. the male was removed. On the 4th of June at 8:00 P.M. the eggs that had remained in the uterus of the female were artificially fertilized. They are recorded as 89 hours old at this time. Practically all segmented, and only 5 died at the gastrulation stage. From this lot, 434 eggs hatched. Only 12 deaths occurred later. Three hundred of the tad-poles were examined at the time of or after metamorphosis. Of these, 299 were males, and one was a bilateral hermaphrodite.

There can be little doubt, therefore, that, in some way, delay in fertilization has caused practically all the eggs to produce males; and the evidence is the clearer since the eggs fresh laid, fertilized by the same male, produced 55 males and 53 females.

It may seem futile, therefore, to attempt to explain this result in any other way than as the result of the action of the environment on the sex of the egg. But how has the environment

¹ *Festschrift*, R. Hertwig, Bd. II, 1910.

² *Anatom. Anzeiger*, 1911.

acted? The evidence that sex is regulated by an internal mechanism has become so strong in recent years that until the action of the environment is made clear one may well hesitate to accept the case as showing that sex is actually changed or produced by an external agent. Curiously enough, every one seems to have overlooked still another possibility that may solve the difficulty. The delay in the fertilization may cause the polar spindle to stick to the surface of the egg so that it fails later to take part in the development, in which case the sperm nucleus alone would produce the nuclei of the embryo. Or, on the other hand, the delay may cause the early stages in the formation of the female pronucleus to progress so far that after fertilization the sperm nucleus may be excluded in part or entirely from the development. In either case the presence of a single nucleus would be expected to give rise to a male. It is significant in this connection that the changes described by King that affect the sex-ratio of the frogs' eggs produce a higher percentage of males.

There is another curious fact in relation to sex-determination in the frog. Pflüger described a high percentage of hermaphrodites amongst the tadpoles. Kuschakewitsch has given a detailed account of the development of the hermaphroditic glands. Most or all of these organs are later transformed into testis. In general it may be said that eggs from a pair give either equal numbers of males and females; or a mixture of males, females, and hermaphrodites; or all hermaphrodites (potentially males). It is possible that the pseudo-hermaphroditic condition may be connected with the failure of one of the two pronuclei to take part in the development.

If the explanation that I have suggested is correct we might expect to find evidence in its support from the number of chromosomes in the tadpoles that develop from these late fertilized eggs. This would be expected if it is the male pronucleus that gives rise to the nuclei of the embryo. But if it is the female pronucleus that is responsible for the result, the number of chromosomes in the cells of the embryo might be haploid or diploid depending on whether the second polar body was, or was not given off. At any rate, this suggestion should be put to the test of observation before we conclude that sex may be determined by external agents. If the view here suggested prove true, sex is still determined by an internal factor in the same sense that the sex of the bee's egg is determined by the presence of one or of two pronuclei.

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